GLYCEMIA AND LIPIDEMIA VARIATIONS OF THE RABBITS INOCULATED WITH BEE VENOM

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Abstract

Besides the toxic, inflammatory and allergic effect of the bee venom on the mammals the researches in the last decades made also evident its therapeutic effects. To prevent the dismetabolic disease and their consequences on the human being and animals a great attention is paied to the study on the controlling mechanisms of glycemia and lipidemia. In this connection, the present study made evident the serum level variations of glucose, cholesterol and triglycerides 2, 6 and 24 h after the inoculation of the bee venom to rabbits by applying 3 stings (E1 group), 6 stings (E2 group) and 12 stings (E3 group). The obtained results are indicative of a reducing tendency of glycemia 2 h after inoculation at the E1 group while with the E2 and E3 groups the serum glucose level showed a gradual increase without exceeding the physiological reference limits along the whole experiment (due to both the secretion stimulation of catecholamines and the corticosteroidic effect). The levels of cholesterol and serum triglycerides showed a significant gradual decrease with every experimental group 2 and 6 h after inoculation and the recovery of the initial values after 24 h for E1 and E2 but maintained at lower values than the average of the reference at E3.

Key words: bee venom, sting, rabbit, serum glucose, serum cholesterol, serum triglycerides

INTRODUCTION

The researches reported in recent decades revealed the bee venom to show both toxic, inflammatory, allergic as well as therapeutic effects on mammalians. In order to prevent from the dismetabolic diseases and their consequencies at human beings and animals a great interest to the study on the controlling mechanisms of glycemia and lipidemia [6, 7, 9, 12].

The bee venom is used in the alternative medicine for its effects of reducing the glycemia and cholesterolemia being a remedy accepted for treating the diabetes and the atherosclerosis (C. Neacşu, 2002). However, many studies attest the hyperglycemic effect of the bee venom and others revealed also an increase in the blood cholesterol level [10, 12, 14].

These controversial effects of the bee venom mentioned in scientific literature, made us to initiate the present study to settle if the mammalian organism shows a variation of the response to the bee venom in function of the dose and action time.

MATERIAL AND METHOD

The glycemia, cholesterolemia and triglyceridemia were determined on 18 rabbits divided into 3 groups of 6 animals each. The rabbits of 6-7 months old and weighting 0,700-0,900 kg were chosen. They were inoculated with bee venom by applying a variable number of directed stings as follows: E1 group - 3 stings, E2 group - 6 stings, E3 group - 12 stings (one sting is equivalent to 0.1 mg venom). In our experimental model the progressive dose variant and periodical drawing were applied. Thus blood samples were drawn from the rabbits at the moment T0 (prior to the venom inoculation) T1 (2 h after inoculation), T2 (6 h after inoculation) and T3 (24 h after inoculation).

The blood was drawn from the saphenous vein in dry tubes and the serum resulting by centrifuging was submitted to biochemical investigations in the clinical laboratory of the Faculty of Veterinary Medicine Iasi. The serum concentrations of glucose, cholesterol and triglycerides were measured on the automatic biochemical analyzer Accent 200.

RESULTS AND DISCUSSION

The obtained data by our investigations are presented in Table 1.

With the E1 group (0.3 mg bee venom as individual inoculation) the level of the blood serum glucose decreased non-significantly at the time T1 followed by an increase at T2 and the tendency of recovering at the initial value after 24h. These variations are placed within the reference limits of the species. The cholesterol level decreased progressively within the 2-6 h time interval, from 156.2±5.75 mg/dl prior to the venom inoculation to 95.4±2.35 mg/dl 6 h. later, coming then back to the value of the moment T0 exceeding even this value (160.4±10.30 mg/dl). The same progressive decrease at 2-6 h. after inoculation was noticed for the triglyceride serum level followed by a recovery to the normal value after 24 h.

By taking the average value of the standard group at the moment T0 (105.88 mg/dl) as a reference the glycemia level was noticed to increase starting with the moment T2 (6 h after inoculation) – 125.9±2.05 mg/dl, the reference upper limit of the species being not exceeded In comparison with the average value of the control group at the moment T0 (120.03), the serum cholesterol level decreased to 95.4±2.35 mg/dl 6 h. after inoculation with no recovery to the reference limits given in scientific literature (10-80 mg/dl). Triglyceridemia decreased under the bee venom action from the increased values of 198.4±20.30 mg/dl to 71.5±3.65 mg/dl, below the average value of the control group (143.1mg/dl) and of the reference minimum limit of the species. (124-156 mg/dl).

With the E2 group (6 bee stings, 0.6 mg venom per rabbit), the glycemia level increased just after inoculation at the moment T1 increasing then progressively to the value of 129.5±10.4 mg/dl after 24 h. The increase exceeded the average value of the standard group (105.88 mg/dl) with no exceeding of the reference upper limit of the species (155 mg/dl). The cholesterolemia decreased progressively from medium values of 93.0±1.40 mg/dl at the moment T0 to medium values of 37.2±6.95 mg/dl at the time T2 with a recovery tendency to the initial value after 24 h. The level of serum triglycerides shows the same significant progressive diminution from 122.6±25.92 mg/dl at T0 to 37.6±9.95 mg/dl at T2 followed by an increase above the initial value after 24 h. These modifications of lipidemia starting from pathologically increased values make evident a clear hypocholesterolemic and hypotriglyceridemic effect of the dose of 0.6 mg venom per animal occurring immediately after inoculation and being maintained at least 6 h after inoculation

With the E3 group (12 bee stings, 1.2 mg venom per rabbit), the glycemia increase is transitory at the moment T1 only, recovering then to the initial values even from T2. The evolution of cholesterolemia and triglyceridemia is similar to that of the E2 group.

The glycemia variations with all groups under study lie within the reference physiological limits the standard group at T0 moment among them, while the variations of the lipid metabolism parameters are extremely oscillatory both within and beyond the reference physiological limits of the species even at the T0 experimental time. This aspect allows to point out the hypocholesterolemic and hypo-triglyceridemic effects stronger at the groups where the levels of cholesterol and serum triglycerides exceed the reference upper limit of the species.

In scientific literature the cellular cytotoxic effects of the peptides and enzymes in the bee venom are mentioned as well as those on the glycemia and cholesterolemia and the involvement of some components of the bee venom in the mechanisms of intracellular transport of glucose and other metabolites [1, 2, 3, 4, 8, 12].

Table 1. Dynamics of glycemia, cholesterolemia and triglyceridemia (average values and standard deviations) at rabbits inoculated with various bee venom doses (E1 group - 3 stings, E2 group - 6 stings, E3 group - 12 stings; M - control group)

Group	Drawing moment (T)	Measured biochemical parameters		
		Cholesterol mg/dl	Triglycerides mg/dl	Glucose Mg/dl
E1 (n = 6)	T0	156.2 ± 5.75	198.4 ±20.30	103.3 ±7.11
	T1	112.9 ±5.10	76.8 ± 14.15	100.2 ±18.52
	T2	95.4 ±2.35	71.5 ±3.65	125.9 ±2.05
	T3	160.4 ±10.30	129.8 ±9.50	114.3 ±6.15
E2 (n = 6)	T0	93.0 ± 1.40	122.6 ±21.95	96.75 ±3.04
	T1	57.9 ±4.30	64.1 ±3.20	117.2 ±5.95
	T2	37.2 ±6.95	37.6 ±9.95	122.7 ±3.00
	T3	74.8 ±24.30	144.25 ±17.25	129.5 ±10.4
E3 (n = 6)	T0	110.9 ±25.10	108.5 ±21.20	117.6 ±17.20
	T1	78.6 ±20.30	72.5 ±10.45	127.6 ±9.90
	T2	51.9 ±13.25	34.0 ±4.00	116.14 ±9.62
	T3	91.9 ±25.10	85.9 ±42.05	123.73 ±2.64
M (n = 18)		*120.03*	*143.1*	*105.88*
Reference values ([a]Merck, 2008; [b]Marcu Elena, 1999)		***10-80[a]***	***124-156[b]***	***75-155[a]***

T0 - before inoculation; T1 - 2 hours after inoculation; T2 - 6 hours after inoculation; T3 – 24 hours after inoculation.

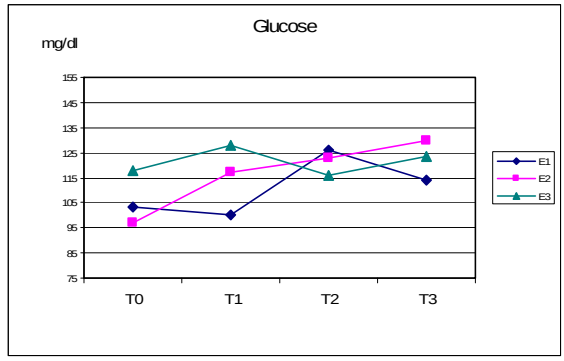

Fig.1. Variation of the serum level of glucose at rabbits inoculated with bee venom in function of dose and the moment of sample drawing (group E1-0.3 mg individual dose; group E2-0.6 mg individual dose; group E3-1.2 mg individual dose. T0-before inoculation; T1-2h after inoculation; T2-6h after inoculation; T3-24h after inoculation).

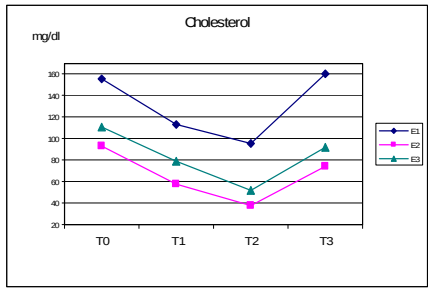

Fig.2. Variation of the serum level of cholesterol at rabbits inoculated with bee venom in function of dose and the moment of sample drawing (group E1-0.3 mg individual dose; group E2-0.6 mg individual dose; group E3-1.2 mg individual dose. T0-before inoculation; T1-2h after inoculation; T2-6h. after inoculation; T3-24h after inoculation).

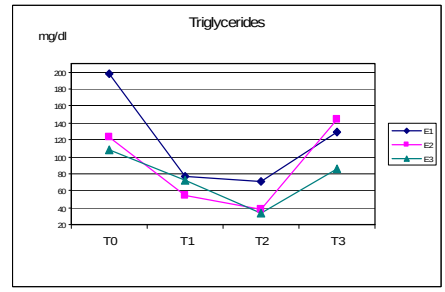

Fig.3. Variation of the serum level of triglycerides at rabbits inoculated with bee venom in function of dose and the moment of sample drawing (group E1-0.3 mg individual dose; group E2-0.6 mg individual dose; group E3-1.2 mg individual dose. T0-before inoculation; T1-2h after inoculation; T2-6h. after inoculation; T3-24h after inoculation).

Thus, the injected bee venom caused the increase in the blood glucose level in rabbit due to the stimulation of the adrenaline secretion and gluconeogenesis increasing. Ishay et al., 1977, injected non-lethal bee venom doses to cat and obtained hyperglycemia and also variations of Na, Ca, K and P concentrations which could be counteracted by propanolol administering [5, 14].

Although in many studies the fosfolipase A2 in bee venom is shown to cause the diminution of the glucose transport rate through the erythrocyte membrane which could explain the restricted increases in the blood glucose level other studies point out the role of A2 phospholipase for intracellular glycolysis increasing by the release of the fatty acids from the membranary phospholipids which activates 6-fosfofructokinase [2, 8].

Han H.J. et al., 2002, demonstrated that the bee venom prevails the glucose from being taken through the apical membrane of the epithelium cells in the proximal renal tube causing thus a shortage in tubular glucose re-absorption at renal level and consequently glucose elimination through urine and decrease in the blood glucose level [3].

Since an autoimmune mechanism is known to act in destroying the beta pancreatic cells, J.Y. Kim et al., 1999, administered to NOD mice (non obese diabetic) 0,5 mg bee venom twice in a week for 4 weeks and noticed an insulin decrease and thus of the diabetes incidence with this experimental model. Another study pointed out that the venom of insects contains substances stimulating the insulin secretion from the pancreatic beta cells of human beings and rats which recommend the venom in diabetes therapy [6, 11].

The most researchers have shown as in our study the decreasing of the serum cholesterol level under the action of bee venom. In other few studies the hypocholesterolemic effect of the insect venom on cats and rats due to the hepatotoxicity promotion is mentioned [10].

The lipoproteins (HDL, LDL, VLDL and chilomicrones) are known to be modified by two plasmatic enzymes: lecithin-cholesterol acyltransferase, with A2 phospholipasic activity and lipoproteinlipase. The action specificity of the two enzymes is the key for the lipid metabolism understanding. The A2 phospholipases in venoms were demonstrated to show enzymatic activity three times higher than that of the plasmatic lecithin-cholesterol acyltransferase [7].

Some authors proved that the A2 phospholipase in the bee venom shows a higher affinity to the plasmatic lipoproteins than to membranary phosphatidylcholines and exerts its cytotoxic effect mostly by generating free fatty acids and lisophospholipids. The free cholesterol in HDL is esterified by the phospholipase

activity (C. Guillaume, 2006). However, the partial lysis of the membranary phosphatidylcholine in adypocites by A2 phosphofolipase in venom affords the binding of a greater number of insulin molecules promoting thus an increase in the glucose transport as well as an acceleration of taking the lipids in the adipose tissue [8].

These effects generated by the A2 phospholipase in the bee venom can explain the hypocholesterolemic and hypotriglyceridemic effect unanimously accepted and made evident in the present study with both low and high venom doses.

CONCLUSIONS

1. By administering bee venom to rabbits a glycemia increase was noticed (with no exceeding the reference limits of the species) with doses of 0.3 mg per animal, beginning with 6h after inoculation and doses of 0.6 mg and 1.2 mg per animal, 2 h after inoculation with recovering the initial values after 24 h. The nonsignificant and transitory increase of glycemia is due to the adrenergic and corticosteroidic effect.

2. The A2 phospholipase in the bee venom determined o significant reduction of cholesterolemia and triglyceridemia at the rabbits under study, significant at 2 and 6 h from inoculation and recovering the initial values after 24h. The effect is the overall result of the enzymatic action on the plasmatic lipoproteins and the taking of triglycerides in the adipose tissue.

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